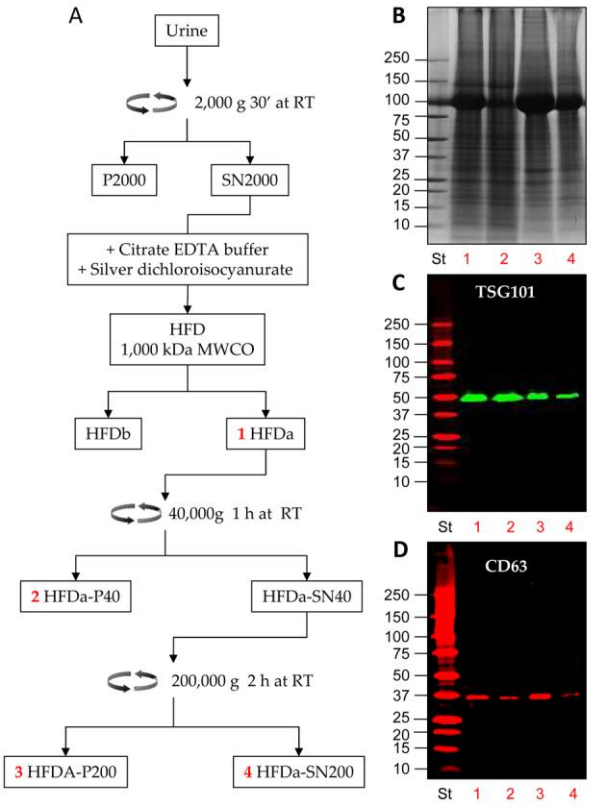
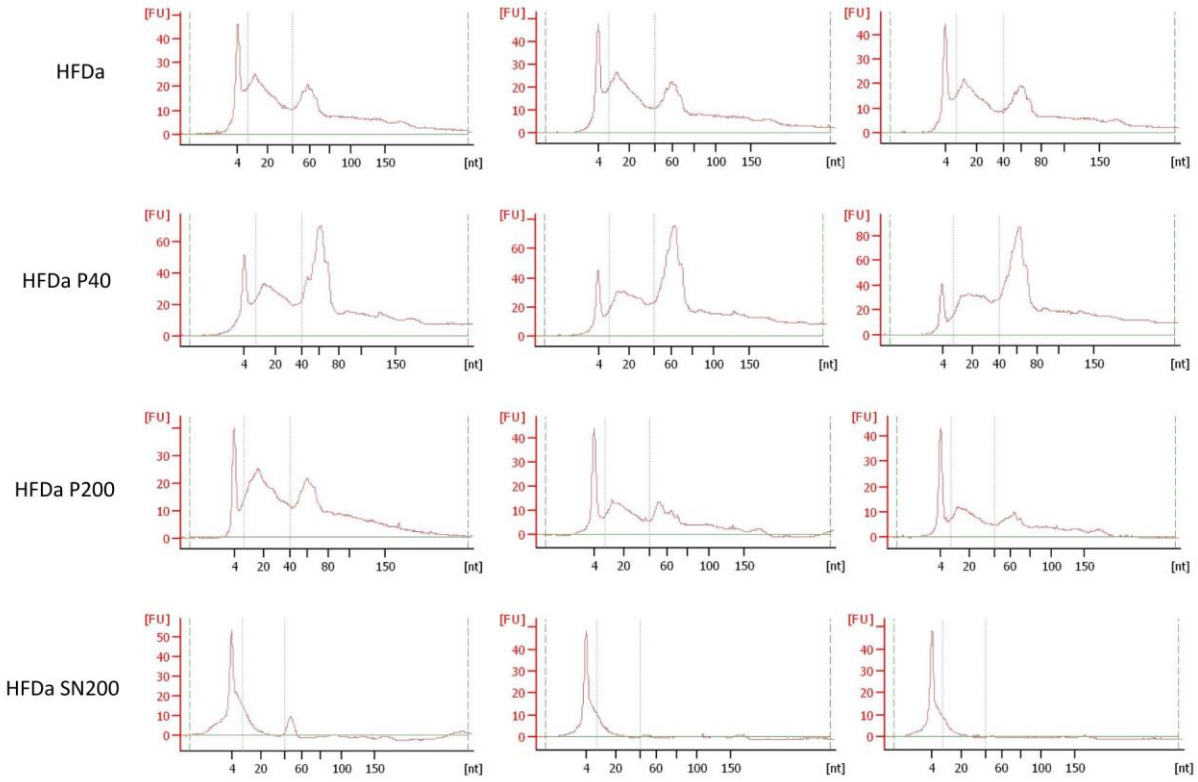
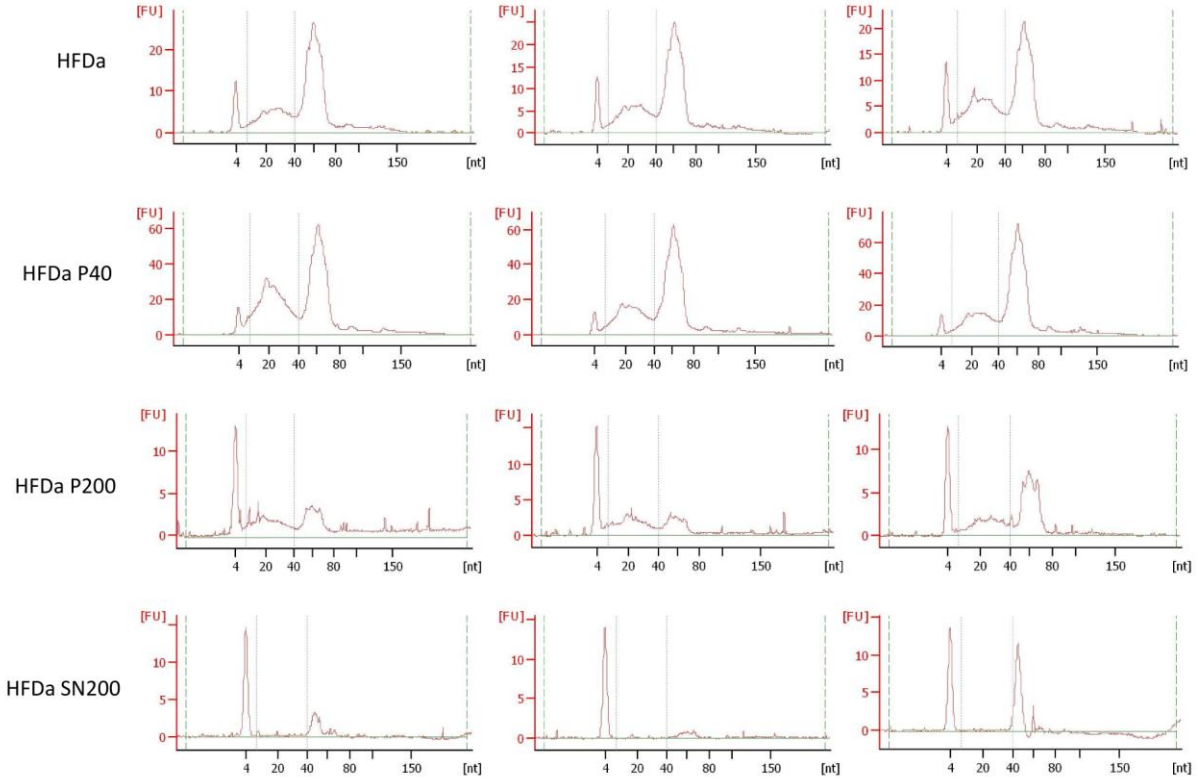


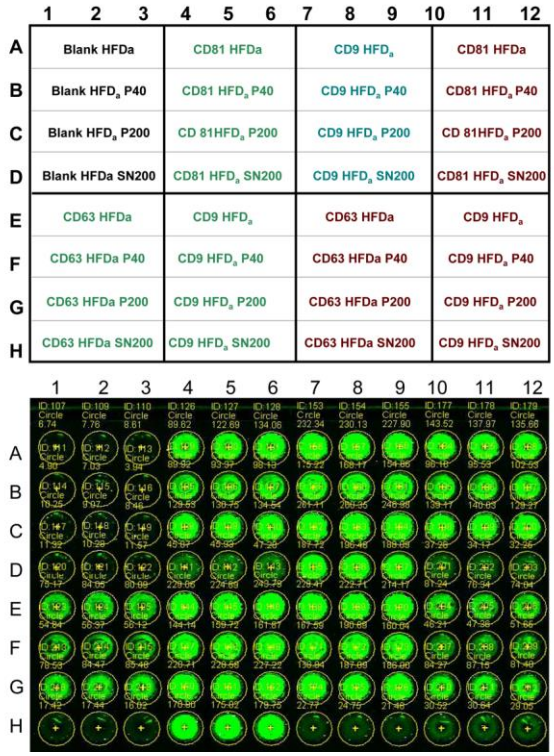
Supplemental Figure S1: **Urinary vesicle enrichment, fractionation and vesicle marker detection.** Panel A**:** workflow to isolate and fractionate urinary vesicles by hydrostatic filtration dialysis followed by differential centrifugation. Urine SN2000 g (1.25 litres) was poured in 1 l separating funnel, after concentration 200 ml of milliQ water was added to flush away all the analytes smaller than the MWCO. HFDa was ready for the differential centrifugation in approximately 24 hours meanwhile the differential centrifugation protocol required nearly half working day. Panel B: Coomassie gel of all vesicles fractions isolated following the workflow (numbered in red). Panel C and D: immunodetection of TSG101 and CD63 (R & D System Antibody). Ten µg of protein (Bradford assay) was loaded per lane. Lane 1 HFDa; Lane 2 HFDa-P40; Lane 3 HFDa-P200, Lane 4 HFDa-SN200. St Molecular weight standard expressed in kDa



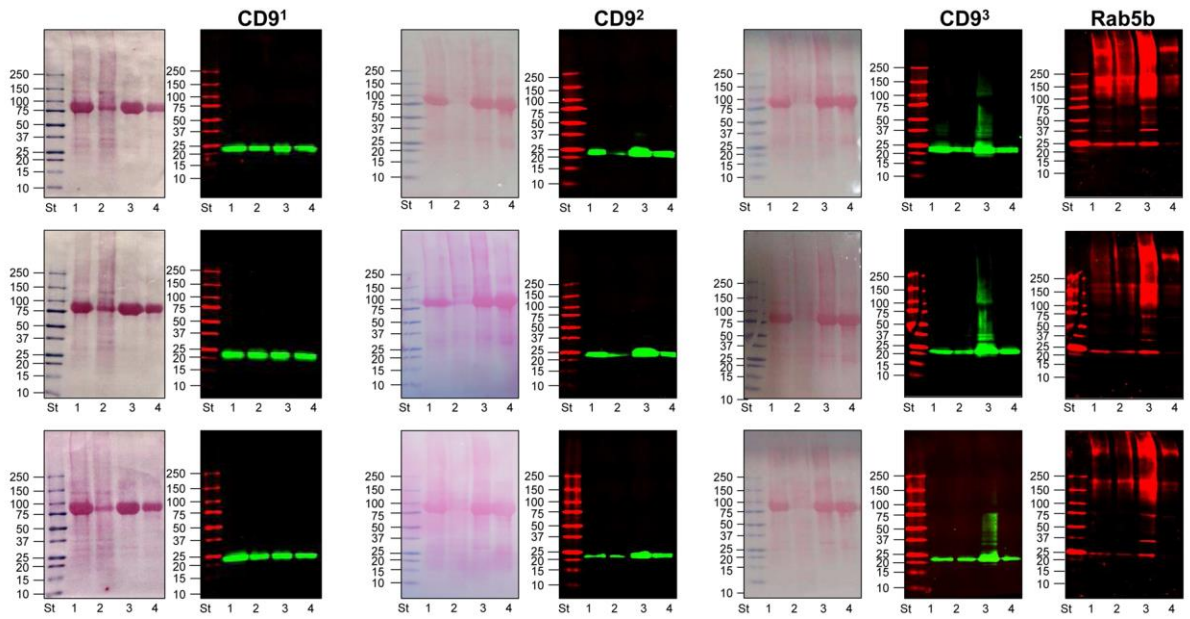
Supplemental Figure S2. Isolated RNA profiles of triplicate extraction using Norgen isolation Kit. Electropherograms of 1 µl of RNA extracted from HFDa, HFDa-P40, HFDa-P200 and HFDa-SN200 were determined using small RNA Chip



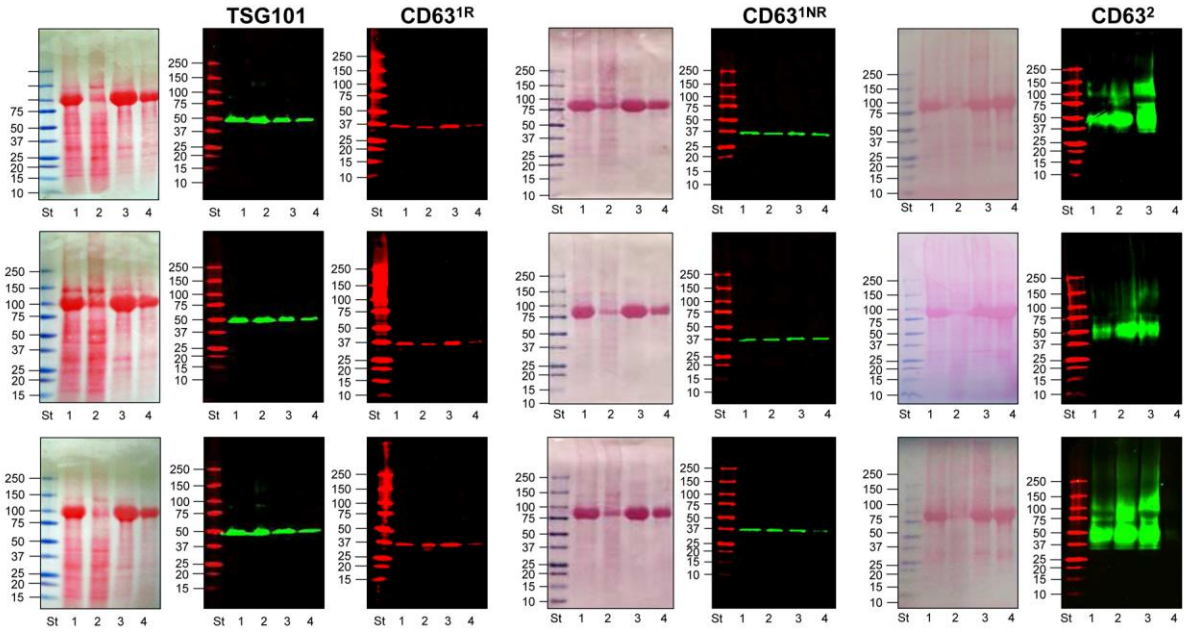
Supplemental Figure S3. Isolated RNA profiles of triplicate extraction using mirVana™ extraction Kit. Electropherograms of 1 µl of RNA extracted from HFDa, HFDA-P40, HFDa-P200 and HFDa-SN200 were determined using small RNA Chip



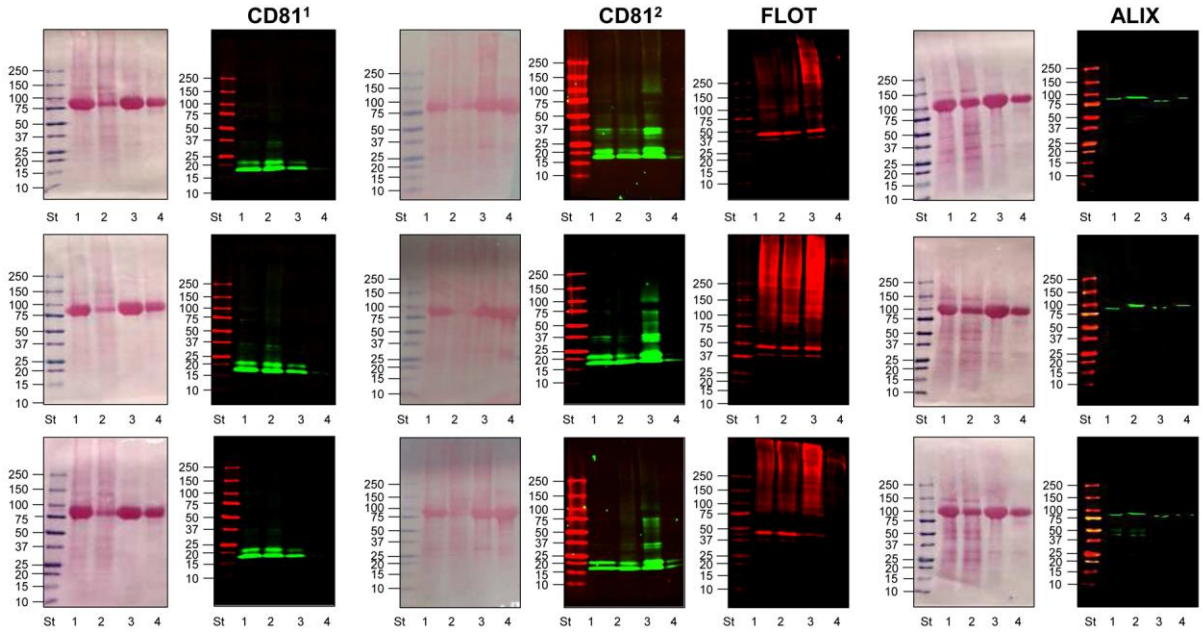
Supplemental Figure S4: Fluorophore-linked immunoasorbent assay (FLISA). Ten μg of protein vesicles immobilized FLISA per each fraction obtained from the differential centrifugation protocol were used to coat the well. A) CD9 (green R & D antibody, blue Merck Millipore antibody, dark red HansaBioMed antibody); B) CD81; (green R & D antibody, red HansaBioMed antibody); C) CD63 (green R & D antibody, red HansaBioMed antibody)



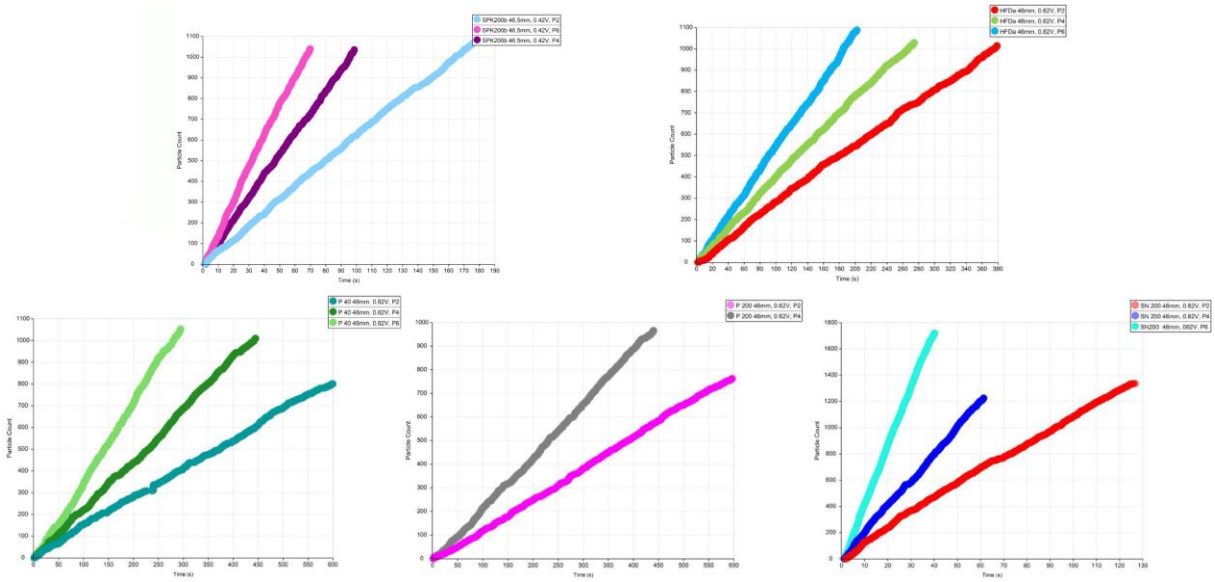
Supplemental Figure S5: Ponceau staining and immunoblots detection for the relarive quantification of vesicle markers CD9 and Rab5 in HFDa and differential centrifugation pellets and supernatant. Gel loading was based on the Bradford assay (10µg of protein per lane). Lane 1 HFDa, Lane 2 HFDa-P40,000 g Lane 3 HFDa-P200,000 g, Lane 4 HFDa-SN200,000 g. Samples were denaturated Samples were denaturated without any DTT. St – molecular weight markers (LI-COR Biosciences) are expressed in kiloDalton. SN-Supernatant; P-Pellet. CD9[1] Mouse anti CD9 R & D System; CD9[2] Mouse anti CD9 Merck Millipore; CD9[3] Mouse anti CD9 HansaBioMed; Rab5 Rabbit anti Ras-relted protein Rab5 HansaBioMed;



Supplemental Figure S6: Ponceau staining and immunoblots detection for the relarive quantification of vesicle markers TSG101 and CD63 in HFDa and differential centrifugation pellets and supernatant. Gel loading was based on the Bradford assay (10µg of protein per lane). Lane 1 HFDa, Lane 2 HFDa-P40,000 g Lane 3 HFDa-P200,000 g, Lane 4 HFDa-SN200,000 g. Samples were denaturated without any DTT for CD63 while 50 mM DTT was added to denaturation buffer for TSG101 St: molecular weight markers (LI-COR Biosciences) are expressed in kilo Dalton. SN-Supernatant; P-Pellet R reducing (+DTT), NR nonreducing, (-DTT),. CD63^{1R} and CD63^{1NR} Mouse anti CD63 R & D System; CD63^{2} Mouse anti CD63 HansaBioMed; TSG101 Rabbit anti tumour susceptibility gene 101Sigma Aldrich



Supplemental Figure 7: Ponceau staining and immunoblots detection for the relarive quantification of vesicle markers CD81, Flotilin 1 and Alix in HFDa and differential centrifugation pellets and supernatant. Gel loading was based on the Bradford assay (10 µg of protein per lane). Lane 1 HFDa, Lane 2 HFDa-P40,000 g Lane 3 HFDa-P200,000 g, Lane 4 HFDa-SN200,000g. Samples were denaturated without any DTT for the detection of CD81 and FLOT 1 while 50 mM DTT was added to denaturation buffer for ALIX. St: molecular weight markers (LI-COR Biosciences) are expressed in kiloDalton. SN-Supernatant; P-Pellet R. CD81[1] Mouse anti CD81 R & D System; CD81[2] Mouse anti CD81 HansaBioMed; FLOT 1 Rabbit anti HansaBioMed, ALIX mouse anti anti-programmed cell death interacting protein Thermo Scientific



Supplemental Figure 8: Particle count rate of particle standard, HFDa and differential centrifugation pellets and supernatant. Particle count-plot illustrates the linearity of the particle detection and count. When extended interruptions and fluctuations in the rate of particle detection occurred the measure was stopped and restarted from the beginning as soon the baseline stabilised again and count ratio resumed